



IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Patent Application of:
Lan Kluwe

Application No.: 10/692,537

Confirmation No.: 9877

Filed: October 24, 2003

Art Unit: 1637

For: METHOD FOR THE DETERMINATION OF
DATA FOR THE PREPARATION OF THE
DIAGNOSIS OF PHAKOMATOSIS

Examiner: Kim, Young J.

MS AF
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Declaration Under 37 C.F.R. §1.132 of Lan Kluwe

Sir:

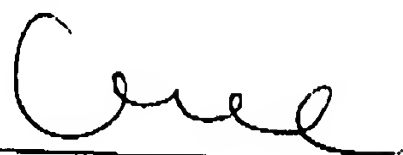
I, Dr. Lan Kluwe of Hamburg, Germany, hereby declare as follows:

1. I am currently employed by the Laboratory for Tumor Biology and Developmental Disorders, Department of Maxillofacial Surgery, of the University Hospital Eppendorf as Laboratory Head. I have been working and studying in the area of phakomatoses since 1993. I received my doctorate degree in Molecular Biology in 1990. A copy of my resume is attached.
2. I am the inventor on the patent application identified above. I have read an Office Action dated April 17, 2007 relating to this application. I understand that the Examiner is of the view that at the June 27, 2000 filing date of this application, it would have been obvious to determine if an offspring of a patient suffering from a phakomatosis tumor suppressor gene disease is at risk of developing the disease using the method as claimed in my patent application.
3. As of the June 27, 2000 original filing date of this application, even though loss of heterozygosity determinations generally were known, I am not aware that anyone was using this method to determine the risk for offspring. In fact, to my knowledge, at the time my application was filed this testing was done by mutation analysis and occasionally by linkage analysis. Linkage analysis, however, can only be applied to families with at least two affected individuals.
4. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements are made with knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title XVIII of the United

States Code and that willful false statements may jeopardize the validity of this Application for Patent or any patent issuing thereon.

Lan Kluwe, Ph.D.

Dated: Oct. 15, 2007

Signature: 

PERSONAL INFORMATION:

Name: Lan Kluwe
Gender: Female
Nationality: German
Occupation: Head of the Laboratory for Tumor Biology and Developmental Disorders,
Department of Maxillofacial Surgery, University Hospital Eppendorf,
Martinistr. 52, 20246 Hamburg, Germany.
Contact information: phone +49-40-42803-8267; Fax +49-40-42803-9665
kluwe@uke.uni-hamburg.de

EDUCATION:

09.1979 - 02.1980 Undergraduate school at the Dept. of Psychology, Beijing University,
Beijing, China.
04.1981-02.1985 Undergraduate school at the Dept. of Biology, Tohoku University, Sendai,
Japan. **Bachelor** of Biology
04.1985-03.1990 Graduate school at the Dept of Molecular Biology, Nagoya University,
Nagoya, Japan. **Ph.D.**

OCCUPATION:

06.1990-04.1993 Post Doc at European Molecular Biology Laboratory, Outstation Hamburg at
Deutsche Synchrotron.
05.1993-06.1993 Invited researcher at the Dept of Biochemistry, Birmingham University,
Birmingham, England
07.1993-present Director, Laboratory for phacomartosis and malformation, Department of
Maxillofacial Surgery, University Hospital Eppendorf, Hamburg, Germany.
10.2006-present Invited senior researcher and project leader in the Neurogenetic Center,
Massachusetts General Hospital, Boston, US

PUBLICATIONS (see detailed list)

First author: 23 (in Mol Hum Genet, Hum Mutat, J Med Genet, FEBS Lett, J Bacteriol, ect.)
Last author: 6
Co-autor: 38

Publikationen

First author

1. **Lee L**, Mizuno T, Imae Y: Thermosensing properties of Escherichia coli tsr mutants defective in serine chemoreception. J Bacteriol 1988;170:4769-4774
2. **Lee L**, Imae Y: Role of threonine residue 154 in receptor function of the Tar chemoreceptor in Escherichia coli. J Bacteriol 1990;172:377-382
3. **Kluwe L**, Maeda K, Maeda Y: E.coli expression and characterization of a mutant troponin I with the three cysteine residues substituted. FEBS 1993;Lett.323:83-88
4. **Kluwe L**, Maeda K, Miegel A, Fujita-Becker S, Maeda Y, Talbo G, Houthaeve T, Kellner R: Rabbit skeletal muscle alpha-tropomyosin expressed in baculovirus-infected insect cells possesses the authentic N-terminus structure and function. J Muscle Res Cell Motil 1995;16:103-110
5. **Kluwe L**, Pulst SM, Mautner VF: A 163-bp deletion in the neurofibromatosis 2 (NF2) gene associated with variant phenotypes. Hum Genet 1995;95:443-446
6. **Kluwe L**, Mautner VF: A missense mutation in the NF2 gene results in moderate and mild clinical phenotypes of neurofibromatosis type 2. Hum Genet 1996;97:224-227
7. **Kluwe L**, Bayer S, Baser ME, Hazim W, Haase W, Funsterer C, Mautner VF: Identification of NF2 germline mutations and comparison with neurofibromatosis 2 phenotypes. Hum Genet 1996;98:534-8
8. **Kluwe L**, MacCollin M, Tatagiba M, Thomas S, Hazim W, Haase W, Mautner VF: Phenotypic variability associated with 14 splice-site mutations in the NF2 gene. Am J Med Genet 1998;77:228-33
9. **Kluwe L**, Mautner VF: Mosaicism in sporadic neurofibromatosis 2 patients. Hum Mol Genet 1998;7:2051-2055
10. **Kluwe L**, Friedrich R, Mautner VF: Loss of NF1 alleles in Schwann cells but not in fibroblasts derived from a NF1-associated neurofibroma. Genes Chromosom Cancer 1998; 24:283-285
11. **Kluwe L**, Friedrich R, Mautner VF: Allelic loss of the NF1 Gene in NF1-associated plexiform neurofibromas. Cytogenet Cancer Genet 1999;110:103-10
12. **Kluwe L**, Friedrich RE, Hagel C, Lindenau M, Mautner V-F: Mutations and Allelic loss of the NF2 gene in neurofibromatosis 2-associated skin tumors. J Invest Dermatol 2000; 114:1017-1021
13. **Kluwe L**, Mautner VF, Parry DM, Jacoby LB, Baser M, Gusella J, Davis K, Stavrou D, MacCollin M: Parental origin of de novo mutations in sporadic neurofibromatosis 2 patients 2000, Neurogenet 2000;3:17-24.
14. **Kluwe L**, Hagel C, Tatagiba M, Thomas S, Stavrou D, Ostertag H, von Deimling A, Mautner VF: Loss of NF1 alleles distinguish sporadic from NF1-associated pilocytic astrocytomas. J Neuropathol Exp Neurol. 2001;60:917-20.
15. **Kluwe L**, Friedrich RE, Tatagiba M, Mautner VF: Presymptomatic diagnosis for children of sporadic neurofibromatosis 2 patients: A method based on tumor analysis. Genet Med. 2002 Jan-Feb;4(1):27-30.
16. **Kluwe L**, Friedrich RE, Korf B, Fahsold R, Mautner VF: NF1 mutations in neurofibromatosis 1 patients with plexiform neurofibromas. Hum Mutat. 2002 Mar;19(3):309.
17. **Kluwe L**, Mautner V, Heinrich B, Dezube R, Jacoby LB, Friedrich RE, MacCollin M. 2003. Molecular study of frequency of mosaicism in neurofibromatosis 2 patients with bilateral vestibular schwannomas. J Med Genet 40(2):109-114.
18. **Kluwe L**, Friedrich RE, Peiper M, Friedman J, Mautner VF. 2003. Constitutional NF1 mutations in neurofibromatosis 1 patients with malignant peripheral nerve sheath tumors. Hum Mutat 22(5):420.
19. **Kluwe L**, Tatagiba M, Funsterer C, Mautner VF. 2003. NF1 mutations and clinical spectrum in patients with spinal neurofibromas. J Med Genet 40(5):368-371.
20. **Kluwe L**, Mautner V, Heinrich B, Dezube R, Jacoby LB, Friedrich RE, MacCollin M. 2003. Molecular study of frequency of mosaicism in neurofibromatosis 2 patients with bilateral vestibular schwannomas. J Med Genet 40(2):109-114.
21. **Kluwe L**, Siebert R, Gesk S, Friedrich RE, Tinschert S, Kehrer-Sawatzki H, Mautner VF. 2004. Screening 500 unselected neurofibromatosis 1 patients for deletions of the NF1 gene. Hum Mutat 23(2):111-116.
- Kluwe L**, Nygren AO, Errami A, Heinrich B, Matthies C, Tatagiba M, Mautner V. 2005. Screening for large mutations of the NF2 gene. Genes Chromosomes Cancer 42(4):384-391.
22. **Kluwe L**. 2006. Schwannomatosis. In Atlas of Genetics and Cytogenetics in Oncology and Haematology (<http://www.infobiogen.fr/services/chromcancer/>)
23. **Kluwe L, Hagel H, Mautner V**. 2007. Neurofibromas. In Atlas of Genetics and Cytogenetics in Oncology and Haematology (in press).
24. **Kluwe L**. 2007. Molecular studies on schwannomatosis. In Nanographies in Human Genetics. Vol. 16: Neurofibromatoses. Karger Press (in press)
25. **Kluwe L, Hagel C, Mautner V**. Neurofibroma. Atlas Genet Cytogenet Oncol Haematol. April 2007 . URL : <http://AtlasGeneticsOncology.org/Tumors/NeurofibromaID5098.html>

Last author

- 1 Mautner VF, Baser ME, **Kluwe L**: Phenotype variability in two families with novel point and frameshift NF2 mutations. Hum Genet 1996;98:203-6
- 2 Lamszus K, Vahldiek F, Mautner V-F, Schichor C, Tonn J, Stavrou D, Fillbrandt R, Westphal M, **Kluwe L**: Allelic losses in neurofibromatosis 2-associated meningiomas. J Neuropathol Exp Neurol. 2000; 59: 504-512
- 3 Mautner V-F, Lindenau M, Gottesleben A, Goethe G, **Kluwe L**: Supporting evidence of a gene for partial epilepsy on 10q. Neurogenet 2000;3:31-4
- 4 Mautner VF, Baser ME, Thakkar SD, Feigen UM, Friedman JM, **Kluwe L**: Vestibular schwannoma growth in patients with neurofibromatosis Type 2: a longitudinal study. J Neurosurg. 2002 Feb;96(2):223-8.

- 5 Frahm S, Mautner VF, Brems H, Legius E, Debiec-Rychter M, Friedrich RE, Knofel WT, Peiper M, **Kluwe L**. 2004. Genetic and phenotypic characterization of tumor cells derived from malignant peripheral nerve sheath tumors of neurofibromatosis type 1 patients. *Neurobiol Dis* 16(1):85-91.
- 6 Lammert M, Mautner VF, **Kluwe L**. 2005. Do hormonal contraceptives stimulate growth of neurofibromas? A survey on 59 NF1 patients. *BMC Cancer* 5:16.

Co-author

- 1 Matsuoka A, Iwasa H, Takiguchi K, Arakawa N, **Lee L**, Takagi T, Shikama K: Stability Properties of oxymyoglobin from chicken gizzard smooth muscle. *Comp Biochem Physiol* 1987;88B:783-789
- 2 Nara T, **Lee L**, Imae Y: Thermosensing ability of Trg and Tap chemoreceptors in Escherichia coli. *J Bacteriol* 1991;173:1120-1124
- 3 Gomi S, **Lee L**, Iwama T, Imae Y: Inhibition of aspartate chemotaxis of Escherichia coli by site-directed sulfhydryl modification of the receptor. *J Biochem* 1992;113:208-213
- 4 Fujita-Becker S, **Kluwe L**, Miegel A, Maeda K, Maeda Y: Reconstitution of rabbit skeletal muscle troponin from the recombinant subunits all expressed in and purified from E.coli. *J Biochem* 1993;114:438-444
- 5 Miegel A, **Lee L**, Dauter Z, Maeda Y: A new crystal form of tropomyosin. *Adv Exp med Biol* 1993;332:25-32
- 6 Tatsuno I, **Lee L**, Kawagishi I, Homma M, Imae Y: Transmembrane signalling by the chimeric chemosensory receptors of Escherichia coli Tsr and Tar with heterologous membrane-spanning regions. *Mol Microbiol* 1994;14:755-762
- 7 Schröder S, **Kluwe L**, Mautner VF: Nicht klassifizierbare Neurofibromatose. Inkomplette Expression von NF1 oder NF2?. *Verhandlungen der Deutschen Gesellschaft für Neurologie* 1994;8:465-467
- 8 Mautner VF, Lindenau M, Hazim W, Tatagiba M, **Kluwe L**, Köppen J, Pulst SM: Phäno-Genotyp-Analyse der NF2. *Verhandlungen der Deutschen Gesellschaft für Neurologie* 1994;8:462-464
- 9 Mautner VF, Tatagiba M, Lindenau M, Fünsterer C, Pulst SM, **Kluwe L**, Zanella FE: Spinal Tumors in patients with Neurofibromatosis Type 2: MR Imaging Study of Frequency, Multiplicity and Variety. *AJ Radiology* 1995;165:951-955
- 10 Mautner VF, Baser ME, **Kluwe L**: Phenotype variability in two families with novel point and frameshift NF2 mutations. *Hum Genet* 1996;98:203-6
- 11 Strauss JD, Van Eyk JE, Barth Z, **Kluwe L**, Maeda K, Wiesner RJ, Rüegg JC: Recombinant troponin 1 substitution and calcium responsiveness in skinned cardiac muscle. *Pflugers Arch* 1996;431:853-62
- 12 Giese A, **Kluwe L**, Laube B, Meissner H, Berens ME, Westphal M: Migration of human glioma cells on myelin. *Neurosurgery* 1996;38:755-64
- 13 Rosenbaum C, **Kluwe L**, Mautner VF, Friedrich RE, Muller HW, Hanemann CO: Isolation and characterization of Schwann cells from neurofibromatosis type 2 patients. *Neurobiol Dis* 1998;5:55-64
- 14 Klose A, Robinson N, Gewies A, **Kluwe L**, Kaufmann D, Buske A, Tinschert S, Peters H: Two novel mutations in exons 19a and 20 and a BsaBI [correction of BsaI] polymorphism in a newly characterized intron of the neurofibromatosis type 1 gene. *Hum Genet* 1998;102:367-71
- 15 Wang Y, Hagel C, Hamel W, Müller S, **Kluwe L**, Westphal M: Trk A, B and C are commonly expressed in human astrocytes and astrocytic gliomas but not by human oligodendrocytes and oligodendroglioma. *Acta Neuropathol* 1998; 96:357-364
- 16 Kamleiter M, Hanemann CO, **Kluwe L**, Rosenbaum C, Wosch S, Mautner VF, Müller H-W, Grafe P: Voltage-dependent membrane currents of cultured human neurofibromatosis type 2 Schwann cells. *Glia* 1998; 24:313-322
- 17 Jacoby LB, MacCollin M, Parry D, **Kluwe L**, Lynch J, Jones Deborah, Gusella JF: Allelic expression of the NF2 gene in neurofibromatosis 2 and schwannomatosis. *Neurogenet* 1998; 7:2051-2055
- 18 Lamszus K, **Kluwe L**, Matschke J, Meissner H, Laas R, Westphal M: Allelic losses at 1p, 9q, 10q, 14q and 22q in the progression of aggressive meningiomas and undifferentiated meningeal sarcomas. *Cancer Genet Cyto Genet* 1999; 10:103-10
- 19 Baser ME, **Kluwe L**, Mautner VF: Germ-Line NF2 Mutations and Disease Severity in Neurofibromatosis Type 2 Patients with Retinal Abnormalities. *Am J Hum Genet* 1999 Apr;64(4):1230-1233
- 20 Mayer C, Kamleiter M, Sanchez-Brandelik R, Rosenbaum C, **Kluwe L**, Hanemann CO, Grafe P: Neuroligand-mediated calcium signaling in cultured human schwannoma cells. *J Peripher Nerv Syst* 1999;4(2):99-105
- 21 Hung G, Faudoa R, Baser ME, Xue Z, **Kluwe L**, Slattery W, Brachman D, Lim D: Neurofibromatosis 2 phenotypes and germ-line NF2 mutations determined by an RNA mismatch method and loss of heterozygosity analysis in NF2 schwannomas. *Cancer Genet Cytogenet* 2000; 118: 167-8
- 22 Rosenbaum C, Kamleiter M, Grafe P, **Kluwe L**, Mautner V, Muller HW, Hanemann CO. Enhanced proliferation and potassium conductance of Schwann cells isolated from NF2 schwannomas can be reduced by quinidine. *Neurobiol Dis.* 2000; 7:483-91
- 23 Bruder CE, Hirvela C, Tapia-Paez I, Fransson I, Segraves R, Hamilton G, Zhang XX, Evans DG, Wallace AJ, Baser ME, Zucman-Rossi J, Hergersberg M, Boltshauser E, Papi L, Rouleau GA, Poptodorov G, Jordanova A, Rask-Andersen H, **Kluwe L**, Mautner V, Sainio M, Hung G, Mathiesen T, Moller C, Pulst SM, Harder H, Helberg A, Honda M, Niihara M, Sahlen S, Blennow E, Albertson DG, Pinkel D, Dumanski JP: High resolution deletion analysis of constitutional DNA from neurofibromatosis type 2 (NF2) patients using microarray-CGH. *Hum Mol Genet* 2001;10:271-82.
- 24 Lamszus K, Lachenmayer L, Heinemann U, **Kluwe L**, Finckh U, Hoppner W, Stavrou D, Fillbrandt R, Westphal M: Molecular genetic alterations on chromosomes 11 and 22 in ependymomas. *Int J Cancer* 2001; 91:803-8.

- 25 Hung G, Li X, Faudoa R, Xeu Z, **Kluwe L**, Rhim JS, Slattery W, Lim D: Establishment and characterization of a schwannoma cell line from a patient with neurofibromatosis 2. *Int J Oncol*. 2002 Mar;20(3):475-82.
- 26 Mautner VF, **Kluwe L**, Thakker SD, Lark RA: Treatment of ADHD in neurofibromatosis type 1. *Dev Med Child Neurol*. 2002 Mar;44(3):164-70.
- 27 Morante-Redolat JM, Gorostidi-Pagola A, Piquer-Sirerol S, Saenz A, Poza JJ, Galan J, Gesk S, Sarafidou T, Mautner VF, Binelli S, Staub E, Hinzmann B, French L, Prud'homme JF, Passarelli D, Scannapieco P, Tassinari CA, Avanzini G, Marti-Masso JF, **Kluwe L**, Deloukas P, Moschonas NK, Michelucci R, Siebert R, Nobile C, Perez-Tur J, Lopez de Munain A. Mutations in the LGI1/Epitempin gene on 10q24 cause autosomal dominant lateral temporal epilepsy. *Hum Mol Genet*. 2002 May 1;11(9):1119-28.
- 28 Hagel C, Lindenau M, Lamszus K, **Kluwe L**, Stavrou D, Mautner VF. Polyneuropathy in neurofibromatosis 2: clinical findings, molecular genetics and neuropathological alterations in sural nerve biopsy specimens. *Acta Neuropathol (Berl)*. 2002 Aug;104(2):179-87.
- 29 Zhao Y, Kumar RA, Baser ME, Evans DG, Wallace A, **Kluwe L**, Mautner VF, Parry DM, Rouleau GA, Joe H, Friedman JM. Intafamilial correlation of clinical manifestations in neurofibromatosis 2 (NF2). *Genet Epidemiol*. 2002; 23:245-159
- 30 Leverkus M, **Kluwe L**, Roli EM, Becker G, Brocker EB, Mautner VF, Hamm H. Multiple unilateral schwannomas: segmental neurofibromatosis type 2 or schwannomatosis? *Br J Dermatol*. 2003 Apr;148(4):804-9.
- 31 Frahm S, Kurtz A, **Kluwe L**, Mautner V-F. Sulindac Derivatives Inhibit Cell Growth and Induce Apoptosis in Primary Cells from Malignant Peripheral Nerve Sheath Tumors of NF1-Patients. *Cancer Cell Int* 4/1/4, 2004
- 32 Kurtz A, Lueth M, **Kluwe L**, Zhang T, Foster R, Mautner VF, Hartmann M, Tan DJ, Martuza RL, Friedrich RE, Driever PH, Wong LJ. Somatic mitochondrial DNA mutations in neurofibromatosis type 1-associated tumors. *Mol Cancer Res*. 2004 Aug;2(8):433-41.
- 33 Kehrer-Sawatzki H, **Kluwe L**, Sandig C, Kohn M, Wimmer K, Krammer U, Peyrl A, Jenne DE, Hansmann I, Mautner V-F. High frequency of mosaicism among patients with neurofibromatosis type 1 (NF2) with microdeletions caused by somatic recombination of the JAZ1 gene. *Am J Hum Genet* 75, 2004
- 34 Lammert M, Friedman JM, **Kluwe L**, Mautner V-F. Prevalence of neurofibromatosis 1 in German children at elementary school enrolment. *Arch Dermatol* 141(1):71-4, 2005
- 35 Lammert M, Friedman JM, Roth HJ, Friedrich RE, **Kluwe L**, Atkins D, Schooler T, Mautner VF. Vitamin D deficiency associated with number of neurofibromas in neurofibromatosis 1. *J Med Genet*. 2006 (10):810
- 36 Miller SJ, Rangwala F, Williams J, Ackerman P, Kong S, Jegga AG, Kaiser S, Aronow BJ, Frahm S, **Kluwe L**, Mautner V, Upadhyaya M, Muir D, Wallace M, Hagen J, Quelle DE, Watson MA, Perry A, Gutmann DH, Ratner N. Large-scale molecular comparison of human schwann cells to malignant peripheral nerve sheath tumor cell lines and tissues. *Cancer Res*. 2006 Mar 1;66(5):2584-91.
- 37 Aghi M, **Kluwe L**, Webster MT, Jacoby LB, Barker FG 2nd, Ojemann RG, Mautner VF, MacCollin M. Unilateral vestibular schwannoma with other neurofibromatosis type 2-related tumors: clinical and molecular study of a unique phenotype. *J Neurosurg*. 2006 Feb;104(2):201-7.
- 38 Mautner VF, Hartmann M, **Kluwe L**, Friedrich RE, Funsterer C. MRI growth patterns of plexiform neurofibromas in patients with neurofibromatosis type 1. *Neuroradiology*. 2006 Mar;48(3):160-5. Epub 2006 Jan 24.
- 39 Lammert M, Friedman JM, Roth HJ, Friedrich RE, **Kluwe L**, Atkins D, Schooler T, Mautner VF. Vitamin D deficiency associated with number of neurofibromas in neurofibromatosis 1. *J Med Genet*. 2006 Oct;43(10):810-3.
- 40 Friedrich RE, **Kluwe L**, Funsterer C, Mautner VF. Malignant peripheral nerve sheath tumors (MPNST) in neurofibromatosis type 1(NF1): Diagnostic findings on magnetic resonance images and mutation analysis of the NF1 gene. *Anticancer Res*. 2005 May-Jun;25(3A):1699-702.
- 41 Kim EL, Yoshizato K, **Kluwe L**, Meissner H, Warnecke G, Zapf S, Westphal M, Deppert W, Giese A. Comparative assessment of the functional p53 status in glioma cells. *Anticancer Res*. 2005 25(1A):213-24.
- 42 Kehrer-Sawatzki H, **Kluwe L**, Funsterer C, Mautner VF. Extensively high load of internal tumors determined by whole body MRI scanning in a patient with neurofibromatosis type 1 and a non-LCR-mediated 2-Mb deletion in 17q11.2. *Hum Genet*. 2005 May;116(6):466-75. Epub 2005 Mar 18.
- 43 Hagel C, Zils U, Peiper M, **Kluwe L**, Gotthard S, Friedrich RE, Zurakowski D, von Deimling A, Mautner VF. Histopathology and clinical outcome of NF1-associated vs. sporadic malignant peripheral nerve sheath tumors. *J Neurooncol*. 2006 Nov 17; [Epub ahead of print]
- 44 Raedt TD, Stephens M, Heyns I, Brems H, Thijs D, Messiaen L, Stephens K, Lázaro C, Wimmer K, Kehrer-Sawatzki H, Vidaud D, **Kluwe L**, Marynen P, Legius E. Conservation of hotspots for recombination in low-copy repeats associated with the NF1 microdeletion. *Nat Genet*. 2006 Dec;38(12):1419-23.